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Draft Genome Sequence of *Clostridium botulinum* B2 450 Strain from Wound Botulism in a Drug User in Italy

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Here, we report the draft genome sequence of *Clostridium botulinum* B2 450, responsible for the first reported case of wound botulism in a drug user in Italy.

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Clostridium botulinum is a microorganism able to produce the botulinum neurotoxin (BoNT), a powerful poison that causes botulism, a serious neuroparalytic disease. There are 3 mainly clinical manifestations of botulism: foodborne (ingestion of BoNT-contaminated foods), intestinal (BoNT is produced by intestinal colonization of *C. botulinum*), and wound (*C. botulinum* spores germinate and synthesize BoNT in contaminated wounds) (1). Based on its physiological characteristics, *C. botulinum* is divided into four different groups (I to IV), so phylogenetically different that they can be considered separate species (2, 3). Moreover, other species, such as *Clostridium baratii* and *Clostridium butyricum*, are able to produce botulinum neurotoxins type F and E, respectively. The BoNT is classified by 8 serotypes (A to H), each of which is divided into subtypes. To date, 16 full genomes and several draft assemblies of *C. botulinum* are available, prevalently isolated from food borne and infant cases. A5(B3′)H04402 065 (accession no. FR773526) is the only full genome sequence originating from a wound botulism case (4, 5).

The B2 450 strain was isolated in 2009 from wound exudate of a heroin user patient, in Messina, Sicily, Italy (6). Sequencing was performed using both Roche 454 GS FLX Titanium and Illumina MiSeq platforms. Roche sequencing generated 286,392 single-end reads, with 112,174,234 total sequenced bases, ~28-fold coverage. The reads were *de novo* assembled with the GS Assembler software (Newbler package) into 160 contigs, and 2,430,881 paired-end reads were produced by Illumina sequencing with 731,610,908 total sequenced bases and ~185-fold coverage. Illumina reads were assembled with Abyss-pe producing 497 contigs. 454 and Miseq contigs were combined using Minimus2 software, and 18 contigs were obtained.

A well-known flaw of the 454 platform is the erroneous determination of homopolymer lengths (7); 1,218 homopolymeric stretches were corrected according to Illumina sequences. Nine gaps were closed using 454 and Miseq reads or Sanger sequencing.

The final draft assembly consists of 9 chromosomal contigs, for a total length of 4,070,655 bp, and one plasmidic contig, 250,014 bp long, containing the BoNT/B2 gene. The genome has a G+C content of 27.8%. The 8 remaining gaps are due to repeated sequences (rRNA operon and beta-*N*-acetyl-glucosamidase genes).

The 16S rRNA gene sequence of B2 450 belongs to group I showing 99.8% similarity with the A1 ATCC 3502 16S sequence (NC_009495) (3). Comparing some gene sequences (rpoB-mdh-aroE-hsp60-aceK-oppB-recA) (8), the B2 450 strain appears phylogenetically closer to *Clostridium sporogenes*, with 99.99% similarity to ATCC 15579 (ABKW0200000), than to all other *C. botulinum* strains (96.14% to A1 ATCC 3502 and 95.79% to A3 Loch Maree; NC_010520). *C. sporogenes* phylogenetically belongs to group I (9). The 450 strain may represent a *C. sporogenes* lineage that has recently acquired the BoNT/B gene (perhaps through the plasmid) (10).

Nucleotide sequence accession number. The genome sequence of *C. botulinum* B2 450 was deposited at DDBJ/EMBL/GenBank under the accession no. JXSU00000000. The version described in this paper is the first version.

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